

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:

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Group Art Unit:

Examiner:

Atty. Dkt. No.: 11899.0155.DVUS02
(MOBT:155—3/KAM)

Serial No.:

Filed: August 30, 2001

For: METHOD OF OPTIMIZING SUBSTRATE
POOLS AND BIOSYNTHESIS OF POLY- β -
HYDROXYBUTYRATE-CO-POLY- β -
HYDROXYVALERATE IN BACTERIA AND
PLANTS

PRELIMINARY AMENDMENT

Commissioner for Patents
Washington, D.C. 20231

Sir:

CERTIFICATE OF EXPRESS MAILING

NUMBER **EL521288762US**

DATE OF DEPOSIT: **August 30, 2001**

I hereby certify that this paper or fee is being deposited with the United States Postal Service "EXPRESS MAIL POST OFFICE TO ADDRESSEE" service under 37 C.F.R. 1.10 on the date indicated above and is addressed to: Commissioner for Patents, Washington, DC 20231.


Signature

Please amend this application as follows:

In The Specification

At page 1, line 5, insert the following paragraph:

--This is a divisional of co-pending application Serial No. 09/313,123 filed May 17, 2001, which is a divisional of Serial No. 08/673,388, filed June 28, 1996, now issued (US patent 5,958,745 September 28, 1999), which is a continuation-in-part of Serial No. 08/628,039, filed April 4, 1996, now issued (US patent 5,942,660 August 24, 1999), which is a continuation-in-part of Serial No. 08/614,877, filed March 13, 1996, now issued (US patent 5,959,179 September 28, 1999).--

In the Claims

Cancel claims 1-40, without prejudice.

Please add claims 41- 46 as follows:

41. A threonine deaminase protein which catalyzes the conversion of threonine to α -ketobutyrate, wherein:
 - a. the leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine;
 - b. the leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine, or
 - c. the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine.
42. A nucleic acid sequence encoding a threonine deaminase protein effective to catalyze the conversion of threonine to α -ketobutyrate, wherein:
 - a. the encoded leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine;
 - b. the encoded leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; or

- c. the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine.
43. A recombinant vector comprising a nucleic acid sequence encoding a threonine deaminase protein effective to catalyze the conversion of threonine to α -ketobutyrate, wherein:
- the encoded leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine;
 - the encoded leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; or
 - the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine.
44. A recombinant host cell comprising a nucleic acid sequence encoding a threonine deaminase protein effective to catalyze the conversion of threonine to α -ketobutyrate, wherein:
- the encoded leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine;
 - the encoded leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; or
 - the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine.
45. A method of preparing recombinant host cells useful to convert threonine to α -ketobutyrate, the method comprising:
- selecting a host cell;
 - transforming the selected host cell with a recombinant vector, wherein the recombinant vector comprises a nucleic acid sequence encoding a threonine deaminase protein effective to catalyze the conversion of threonine to α -ketobutyrate, wherein:

the encoded leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; the encoded leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; or the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; and

c. obtaining recombinant host cells.

46. A plant, the genome of which comprises a nucleic acid sequence encoding a threonine deaminase protein effective to catalyze the conversion of threonine to α -ketobutyrate, wherein:

- a. the encoded leucine residue at amino acid position 447 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine;
- b. the encoded leucine residue at amino acid position 481 is replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine; or
- c. the leucine residue at amino acid positions 447 and 481 are independently replaced with alanine, isoleucine, valine, proline, phenylalanine, tryptophan, or methionine.

REMARKS

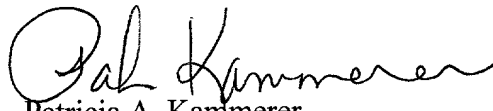
With this amendment, claims 1-40 have been cancelled and claims 41-46 have been added. The active claims in this case are claims 41-46.

The specification has been amended to recite the relationship with the parent case, namely that this application is a divisional of co-pending application Serial No. 09/313,123 filed May 17, 2001, which is a divisional of Serial No. 08/673,388, filed June 28, 1996, now issued (US patent 5,958,745 September 28, 1999), which is a continuation-in-part of Serial No. 08/628,039, filed April 4, 1996, now issued (US patent 5,942,660 August 24, 1999), which is a

continuation-in-part of Serial No. 08/614,877, filed March 13, 1996, now issued (US patent 5,959,179 September 28, 1999).

It is believed that no fee is due; however, should any fees under 37 C.F.R. §§ 1.16 to 1.21 be required for any reason, the Assistant Commissioner is authorized to deduct said fees from our Deposit Account No. 01-2508/11899.0155.DVUS02/KAM.

Respectfully submitted,



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